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Chromatographic Separation and Characterization of Hydrolyzed Cr(III) Species

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Both macroscale and microscale methods to separate hydrolyzed Cr(III) species from acidic to near-neutral pH solutions have been developed. The macroscale approach is based on ion exchange, and involves separating monomeric, dimeric, trimeric, tetrameric, and higher order Cr(III) oligomers from such solutions using a gradient elution with increasing cationic charge. With this approach, the concentration of a given fraction can be maximized, and complete resolution between these species can be achieved. In addition, complete recovery of Cr(III) from the column is achievable. For the microscale approach, capillary electrophoresis with indirect detection is used to isolate and uniquely identify the same smaller oligomers and a fraction of larger Cr(III) species that are not uniquely identified. Capillary electrophoresis also provides indirect structural information for the Cr(III) trimer, suggesting that it exists in a triangular configuration rather than as a linear species. These methods are described in detail, and possible applications are discussed.

The hydrolysis of trivalent chromium has been of interest to inorganic and solution chemists for many decades. In most cases, this interest relates to its use in chemical processes (e.g., the production of stainless steel and other alloys[1−3] and the subsequent waste streams and environmental problems that are generated. Chromium has also been used in casings and structural materials of nuclear fuel packages[1] and as a redox agent in the processing of defense-related nuclear materials.[2] This has led to radioactive wastes contaminated with Cr, actinides, and fission products that must be processed for eventual geologic disposal. This requires that chromium be removed by oxidation to chromate for separation from the radioactivity, but progress in the development of this chemical technology has been hampered by a lack of understanding of Cr(III) speciation and reactivity in the waste systems.[3] Knowledge of the fundamental reactivity of Cr(III) is limited as a result of the complexity of its chemical behavior, the scant structural information about many Cr(III) species, and few analytical tools for effective separation and characterization.

As with other highly charged cations, such as Fe(III), Al(III), and Pu(IV),[4,5] hydrolysis of Cr(III) results in a distribution of oligomeric species formed via μ-hydroxo and μ-oxo bridges between the metal centers that lead to the formation of dimeric, trimeric, tetrameric, and higher order oligomers from the Cr(III) monomer; possible species are shown in Chart 1. Despite decades of study, no agreement exists on the relative importance of these various species in the different chemical processing systems, nor on the actual structures of the Cr(III) trimer and larger oligomers. For example, in a recent paper, we reported on the solubility and speciation of Cr(III) in radioactive waste solutions and found that our experimental solubility data was adequately described by considering only monomeric, dimeric, and possibly trimeric species, even in highly alkaline solutions that should favor the formation of larger Cr(III) oligomers.[6] The coordination geometries shown in Chart 1 for the monomer and dimer are generally accepted for acidic to near-neutral solutions. A search of the literature reveals two possible configurations for the trimer in solution: either linear or triangular, as shown.[7−9] Electron pair resonance data[7] for trimeric Cr(III) in solution did not allow unique identification of either species. Measurement of proton acidities for the μ-hydroxo moieties of the trimer in solution was not consistent with a linear geometry.[9] More recently, we reported structural parameters for the triangular configuration;[10] we were unable to identify a linear Cr(III) species. A crystal structure for

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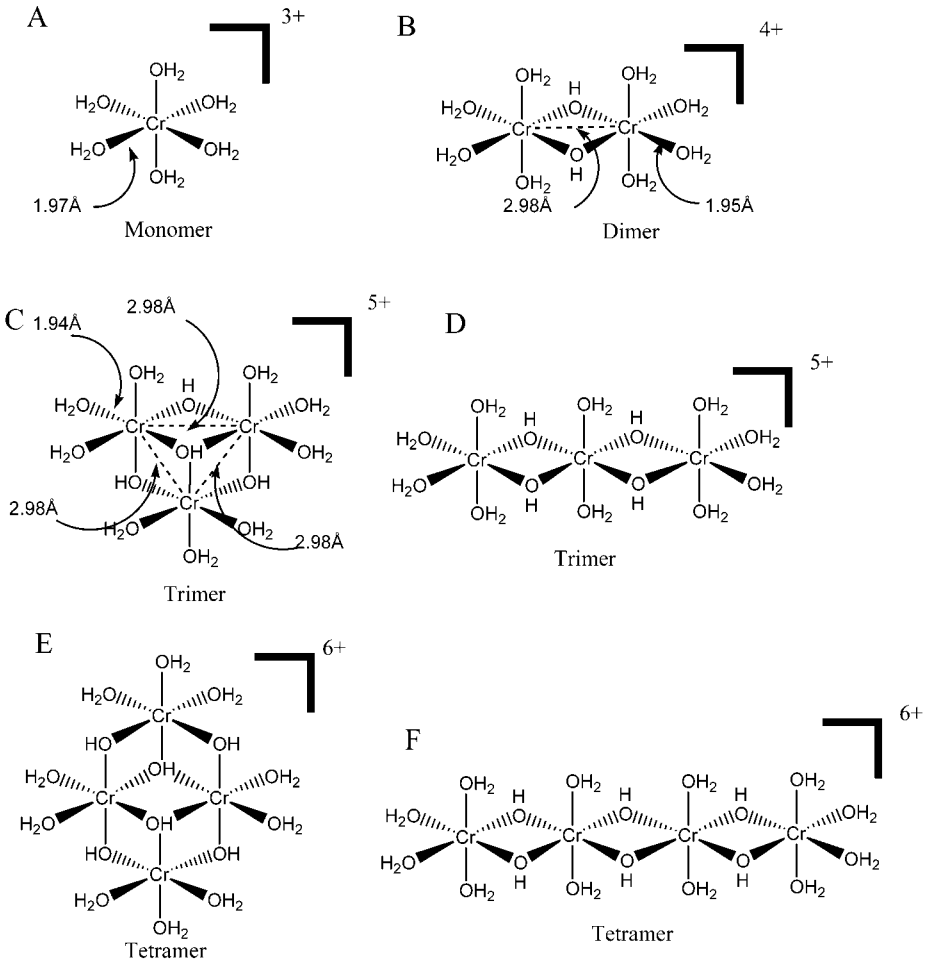
Chart 1. Possible Structures of Solution Cr(III) Species[a]



[a] (A) The hexa-aquo Cr(III) monomer, as suggested by EXAFS;[33] note that only the primary coordination shell is shown. (B) Structure of the Cr(III) dimer based on EXAFS analysis.[16] (C) The triangular configuration for the Cr(III) trimer in solution, as suggested in a recent EXAFS study.[10] (D) The linear conformer for the Cr(III) trimer suggested by electron pair resonance analysis; no structural parameters for this possible species have been reported. (E) The diamond-shaped configuration for the Cr(III) tetramer, as suggested by single-crystal X-ray analysis;[17] no structural parameters for this species in solution have been reported. F. A linear arrangement for the Cr(III) tetramer; this configuration has not been elucidated in either solid phase or solution systems.

the aggregated tetrameric Cr(III) species has been reported,[11] but no structural information for solution species is available. To our knowledge, no firm structural data has been reported for oligomers larger than tetrameric.

In theory, the various species shown in Chart 1 should be easily separable as a result of their different charges. In practice, however, isolation and characterization of the larger Cr(III) oligomers have been problematic because of their large cationic charges distributed over their relatively small structures. Thompson and Connick[7,12,13] reported on the separation of dimeric and trimeric Cr(III) from Cr(III) monomers and higher order oligomers using Dowex 50W-X12 and very large quantities of metal-perchlorate salt solutions as eluants. Although separation was achieved, the work was laborious, and resulting fractions of dimer and trimer were quite dilute. Bradley et al.[14] attempted to separate a very large Cr(III) oligomer using size-exclusion chromatography. They hypothesized the presence of a Cr(III) species analogous to $[AlO_4Al_{12}(OH)_{24}(H_2O)_{12}]^{7+}$ and $[GaO_4Ga_{12}(OH)_{24}(H_2O)_{12}]^{7+}$. Although not confirmed, their work supported the presence of a distribution of large oligomers composed of Cr(III) in an octahedral geometry. No other structural or stoichiometric details were provided. Stunzi et al. have published many reports on the isolation and characterization of monomeric, dimeric, trimeric, and tetrameric Cr(III) species using increasing concentrations of $NaClO_4$ (1.0−4.0 M) in a gradient elution[8,9,15,16] and a Sephadex SP C-25 cation exchanger. Their procedure also required large volumes of eluant that produced dilute fractions of the Cr(III) species.

The dilute fractions of hydrolyzed Cr(III) oligomers isolated by others make spectroscopic characterization and structural determination difficult and often ambiguous. In addition, although rarely mentioned in the literature, these methods leave large quantities of Cr(III) irreversibly sorbed to the ion-exchange or

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size-exclusion resin. This paper describes ion-exchange methods that have been developed to isolate macroscopic quantities of smaller Cr(III) oligomers (e.g., monomer, dimer, trimer, and tetramer) with concentrations that allow further spectroscopic investigation. The method, which relies on a Sephadex cation exchanger and solutions of metal perchlorate salts as eluants, has not (to our knowledge) been reported previously, and is a significant improvement over published methods. As described herein, this method has been demonstrated using both spectroscopic and radiometric techniques. Capillary electrophoresis (CE) is a relatively new chromatographic technique that has been shown to separate microscopic quantities (~ 10 nL) of charged species.[17−21] Separation is based on net species charge density, which is the ratio of overall species charge to its size. CE has been used to isolate minute quantities of larger Cr(III) oligomers, to characterize the purity of the macroscopic fractions of Cr(III) oligomers isolated by ion exchange, and to infer structural information about the Cr(III) trimer on the basis of its net charge density as suggested by CE elution order. Here, these chromatographic methods are described and discussed in terms of further applications for separation of small, highly charged inorganic species and subsequent indirect structural characterization.

EXPERIMENTAL SECTION

Stock solutions of trivalent Cr were made by dissolving $Cr(ClO_4)_3 \cdot 6H_2O$ (Aldrich) in 18 MΩ water to give a deep blue solution. Fresh solutions of base were prepared by dissolution of NaOH or KOH pellets (Fisher). Hydrolysis of the Cr(III) was induced by mixing equal volumes of equimolar concentrations (usually 0.2 M) of Cr(III) with either NaOH or KOH. The base was added dropwise to the Cr(III) solution with rapid stirring. During base addition, the Cr(III) solution color changed from blue to green, becoming increasingly turbid with the formation of a visible precipitate after about 75% of the base had been added. Dropwise base addition was continued until the full volume was consumed. The resulting green suspension was stirred rapidly for 4 h. During this time, the suspended green precipitate gradually dissolved to give a green solution with a pH of ~5.5. At the conclusion of this 4-h period, the pH of the solution was adjusted to 3.0 with 1.0 M $HClO_4$, causing the green color to change to blue-green. This method of hydrolysis, which is a variation of the approach originally reported by Stunzi et al.,[8] was chosen because it is known to yield a large quantity of the smaller Cr(III) oligomers.

In some cases, ^{51}Cr was used as a tracer to monitor the separation of Cr(III) species by ion exchange. ^{51}Cr was produced by neutron irradiation of $Cr(ClO_4)_3 \cdot 6H_2O$ to generate 1.06 μCi of activity. The irradiated $Cr(ClO_4)_3 \cdot 6H_2O$ was dissolved in 18 MΩ water. This solution was green because of small amounts of Cr(III) oxidized to Cr(VI) during irradiation. An aliquot of this solution was then spiked into a nonradioactive Cr(III) solution, which was hydrolyzed as described above to give a solution of 0.069 μCi of ^{51}Cr-labeled oligomers. Appropriate radiation safety procedures must be followed when using tracers such as ^{51}Cr to avoid unnecessary radiation exposure.

Ion Exchange. Ion-exchange columns were 12 cm in length, 1 cm i.d., and filled with 8 cm of Sephadex SP C-25 cation-exchange resin. To prepare the column, the resin was washed with 100 mL of 2 M NaOH, followed by 100 mL of water, and then 100 mL of 2 M HCl. Once the Sephadex was in the proton form, excess protons were removed by a final wash with 100 mL water. Eluants used for elution of the Cr(III) oligomers were $NaClO_4$ (Fisher), $Ca(ClO_4)_2$ (Aldrich), $La(ClO_4)_3$ (Alfa Aesar), $Th(ClO_4)_4$, and $(NH_4)_2Ce(SO_4)_3$. Perchlorate salts, if available, were dissolved directly in 0.01 M $HClO_4$. $Th(NO_3)_4$ (Baker) was dissolved in concentrated $HClO_4$ and fumed to dryness. Dissolution in concentrated perchloric acid followed by fuming was repeated three times. Care must be taken to avoid fuming perchlorate salts in the presence of organics. The excess perchlorate was removed by addition of water and fuming to dryness until the pH upon water dissolution was greater than 1.5. $(NH_4)_2Ce(SO_4)_3$ (Smith Chemical Co.) was dissolved in 0.01 M $HClO_4$ to form a saturated solution.

To isolate a dimer fraction, 3 mL of the hydrolyzed Cr(III) solution was added to the Sephadex column. A portion of 0.5 M $Ca(ClO_4)_2$ was added to wash off the monomer in a clearly visible blue band; a broad blue-greenish band of the dimer also moved down the column at a slower rate. When the dimer band was ~1 cm from the bottom of the column, 0.25 mL of $La(ClO_4)_3$ was added. This caused the dimer band to become more narrow, yielding a small volume (~ 1 mL) of dimeric Cr(III) that was ~30 mM in dimeric species (60 mM in total Cr(III)). Once the dimer was isolated, the top 2 cm of Sephadex, which was deep green, was discarded and replaced with fresh resin prior to column regeneration. Care must be used when handling salts of Th^{4+}, which are all naturally radioactive.

To isolate the trimer, we added 3 mL of hydrolyzed Cr(III) solution, followed by 1.0 M $NaClO_4$ to quickly elute the monomer. Next, 0.5 M $Ca(ClO_4)_2$ was added to elute the dimer as quickly as possible; by this method, the dimer fraction is less concentrated and consequently is discarded. A broad and faint green band of trimer was observed bleeding from the Cr(III) sorbed to the top of the column. When the leading edge of this band was ~1 cm from the end of the column, 0.25 mL of $La(ClO_4)_3$ was added to narrow the band, yielding a 2-mL volume fraction of trimer that was 5−9 mM in trimeric Cr(III) (15−27 mM total Cr(III)). As with the dimer separation, a deep green band remained sorbed to the top of the column; this Cr(III)-laden Sephadex was replaced with fresh resin.

Tetrameric Cr(III) was isolated by first following the method used to isolate the dimer, working as quickly as possible. After the dimer was completely eluted, ~5 mL of $La(ClO_4)_3$ was added to wash off any remaining trimer. With this approach, the trimer band remained broad and poorly defined; it was discarded. To elute a tetrameric fraction from the deep green Cr(III) band sorbed to the top of the column, 0.25 M $Th(ClO_4)_4$ was added. This generated a band that was well-defined, which we eluted in a volume of 3 mL. The fraction was 30 mM in tetramer (120 mM in total Cr(III)). Even with Th^{4+} as the exchanging cation, the top 2 cm of Sephadex remained green. This material was replaced prior to column regeneration.

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Isolated fractions of Cr(III) oligomers obtained by our method were later characterized by a combination of techniques, including inductively coupled plasma atomic emission spectrometry (total Cr concentration), γ spectrometry (^{51}Cr activity levels), capillary electrophoresis (distributions of Cr(III) species and purity of Cr(III) fractions isolated by ion-exchange chromatography), UV−vis spectrophotometry (total Cr and Cr(III) speciation, data reported elsewhere[10]), and by X-ray absorption spectroscopy (structural information and speciation, data reported elsewhere[10]). For those Cr(III) fractions separated by ion exchange that were subsequently analyzed by CE, reduction of the concentrations of simple cations used for elution was necessary prior to CE injection. This was accomplished by treatment with K_2SO_4. The sulfate salts $CaSO_4$, $La_2(SO_4)_3$, and $Th(SO_4)_2$ are all relatively insoluble, as compared to the analogous perchlorate salts, and consequently, are precipitated from solution in the presence of K_2SO_4. In addition, $KClO_4$ also has a relatively low solubility and coprecipitated with the sulfate salts. The Cr(III) oligomers were separated from the precipitates by centrifugation.

Instrumentation. Total Cr concentrations were measured at 205.55 nm using a Jobin Yvon JY24 sequential inductively coupled plasma atomic emission spectrometer equipped with a pneumatic nebulizer. ^{51}Cr activity levels were determined using a Packard Cobra II Auto-gamma counter that utilized a NaI-modified well detector. To count the 320.1 keV g line of ^{51}Cr, energy windows were set between 290 and 400 keV. Samples were counted for 60 min or until 1% RSD was achieved in the overall count rate, whichever came first. UV−vis spectrophotometry was completed using an OLIS-modified Cary 14 spectrophotometer. We have reported electronic spectra and molar absorptivities elsewhere.[10] In general, increasing oligomerization causes a red-shift in absorbance maximums for the Cr(III) species. All measurements were made at room temperature in 1-cm quartz vials.

Capillary Electrophoresis. CE chromatograms were collected using a Dionex capillary electrophoresis instrument employing ultraviolet detection and a Dionex advanced computer interface; Dionex software was used for collection of the raw chromatographic data. The capillary was fused silica, 69 cm long with a 75-mm i.d. Because Cr(III) itself does not have a large molar absorptivity, we used an indirect method of detection, as reported by Chen and Cassidy.[22] For this approach, a buffer consisting of 4 mM α-hydroxyisobutyric acid (HIBA, Aldrich) served as the electrolyte, and 4 mM *N*,*N*-dimethylbenzylamine (DBA, Aldrich) served as the chromaphore. The monochromator of the CE detector was set to 214 nm (molar absorptivity of DBA at $\lambda = 214$ nm is 6.0×10^3 cm^{-1} mol^{-1} L). To this solution matrix, 10 000−30 000 V was applied in positive mode to give ∼2−6 μA of current. The presence of DBA in the matrix gave a large, constant absorbance at 214 nm, unless it was displaced at the point of detection by other nonabsorbing constituents, such as Cr(III) species or other cations. Consequently, these species were registered as a decrease in signal in the electropherograms.

Data Analysis. The Dionex software output is time (in 1.0 s intervals) versus absorbance. Origin 6.0 was used for processing these data, and for peak analysis. The various peaks in the electropherograms were assigned to species using a process of elimination, as described in the Results and Discussion Section. For the simple cations (e.g., Na^+, K^+,Ca^{2+}, La^{3+}, and monomeric Cr^{3+}), the order of elution is consistent with previous reports.[22]

In CE, the order of species elution is determined by overall species charge density, which is the ratio of charge of a cation to its size, including its solvation sphere. In positive mode, the species with the greatest net cationic charge density is detected first. Consequently, the retention time of a cation in the capillary is directly related to its overall size and net positive charge. The hydrated radius (r) of a species can be correlated to its electrophoretic mobility (μ) as follows

$$r = \frac{q}{6\pi\eta\mu} \tag{1}$$

where q is the charge of the species in C and η is the viscosity of the solution in kg m^{-1} s^{-1}. The electrophoretic mobility for a given species is defined as the difference between its apparent mobility (μ_{app}) and the mobility of the electroosmotic flow (μ_{eo}). Both parameters are related to the length of the entire capillary (L_t), length of travel to the detector (L_d), the applied voltage (V), and the retention time (t_n), as follows

$$\mu_{app} = \frac{L_d L_t}{V t_{Cr}}$$

and

$$\mu_{eo} = \frac{L_d L_t}{V t_{EOF}} \tag{2}$$

where t_{Cr} is the retention time of a given Cr species and t_{EOF} is the retention time of the electroosmotic front. Thus, from the retention time for a given Cr(III) species in an electropherogram, information about its overall size is obtained, and information about its shape can be inferred.

RESULTS AND DISCUSSION

In previous work, we showed that aging of Cr(III) species to form larger oligomers is a dynamic process[10] that has not been studied in detail, and little is known about reverse processes. Furthermore, information on the overall net charge densities of these species in solution, and in the case of the trimer, the distribution of this charge within the structure (i.e., overall shape), is limited. We developed ion-exchange and capillary electrophoresis methods to address these problems.

Ion Exchange. Separation of Cr(III) oligomers using previously published methods is difficult. The Dowex ion-exchange method of Finholt, Thompson, and Connick[7,12,13] could not be followed exactly, because the amount of cross-linking for the resin used in their study was no longer commercially available. Isolation of the Cr(III) dimer (which we verified by its electronic transitions[10]) and a second band that was presumably the trimer was accomplished using Dowex 50W-X4 resin. However, the trimer was so dilute that the species could not be characterized by spectroscopic or other methods. Using the Sephadex SP C-25 method reported by Stunzi et al.,[15] isolation of dimeric and trimeric Cr(III) was possible, but overall concentrations of these species were also quite low. In addition, both ion-exchange methods

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Table 1. Recovery of Cr(III) from the Ion Exchange Columns after Completion of the Separations Shown in Figure 1

chromatogram	first wash soln	mole fraction[a] Cr(III) removed in first wash	second wash soln	mole fraction[a] Cr(III) removed in second wash	mole fraction[a] Cr(III) remaining on resin
Figure 1A	5.0 mL, 0.25 M $Th(ClO_4)_4$	0.121 ± 0.004	5.0 mL, 2.0 M HCl	0.122 ± 0.004	0.199 ± 0.002, 21.0 mL of resin
Figure 1B	5.0 mL, 2.0 M $H(ClO_4)_4$	$(7.42 \pm 0.34) \times 10^{-3}$	5.0 mL, 2.0 M $H(ClO_4)_4$	$(0.710 \pm 0.91) \times 10^{-3}$	$(2.70 \pm 0.41) \times 10^{-3}$, 31 mL of resin
Figure 1C	5.0 mL, 2.0 M $H(ClO_4)_4$	$(1.33 \pm 0.51) \times 10^{-3}$	5.0 mL, 2.0 M $H(ClO_4)_4$	$(0.646 \pm 0.95) \times 10^{-3}$	$(6.24 \pm 0.31) \times 10^{-3}$, 24 mL of resin

[a] Mole fractions of Cr(III) are determined from the amount of ^{51}Cr activity either eluted in the wash solutions or remaining on the column.

resulted in very low overall total Cr recoveries from the columns; significant fractions of Cr remained sorbed to the resins at the top of the column. Duplication of the size-exclusion approach reported by Bradley et al.[14] was also attempted. The packed column was prepared with Sephadex G-25 followed by a Sephadex G-10 column with a total length of 75 cm (1-cm i.d.). Blue dextran was run through the column first to determine the void volume, and was quantitatively recovered. Next, a solution of hydrolyzed Cr was passed through the column, but unlike with the ion-exchange columns, no colored bands developed, suggesting little or no separation. In addition, the Cr sorbed to the column, mostly on the top half, and could not be eluted. When blue dextran was run through the same column again, a large fraction of it was retained, apparently as a result of interaction with the sorbed Cr(III).

Ion exchange is best suited for separation of the smaller oligomers in macroscopic amonts. Despite limited success with published methods, they became the basis for our development work. We developed individual approaches to isolate each specific Cr(III) oligomer fraction, as described in the Experimental Section. This was necessary because of reactivity between Cr(III) and the ion-exchange resin and changes in Cr(III) oligomer speciation with time.[23] Each individual separation was optimized to maximize the concentration of the given Cr(III) oligomer for later characterization. Except where noted below, each of these methods leaves large quantities of green Cr(III) sorbed to the top of the column. Visual inspection of the various shades of green, blue, and blue-green bands that developed below the band of green at the top of the column were used to monitor separation on the column itself. Separation and Cr(III) speciation were also monitored by use of the ^{51}Cr tracer. Purity of the separated bands and changes in speciation in those fractions were monitored by CE, as described in detail below.

Using ^{51}Cr as a tracer and a gradient elution approach, a single procedure method was developed to isolate cleanly resolved fractions of these species, as shown in Figure 1. In Figure 1A, the gradient is based on simply increasing cationic charge (e.g., Na^+ to Ca^{2+} to La^{3+} to Th^{4+}), similarly to the individual separations described above, and the chromatographic bands obtained are labeled. Note that although the fractions are generally well separated, the total concentrations of oligomeric species in a given fraction are not necessarily optimized as with the individual separations described in the Experimental Section. In our initial work, a large amount of ^{51}Cr activity remained on the column upon completion of the separations (see Table 1). Even after washing the column with an additional 5 mL of $Th(ClO_4)_4$, which removed ~12% of the Cr, followed by 5 mL of 2.0 M HCl, which removed another 12%, about 20% of the Cr remained "irreversibly" sorbed to the resin.

One of the difficulties of working with $Th(ClO_4)_4$ as an eluant is the presence of natural radioactive daughters, which increases the background counts and decreases our ability to discriminate the chromatographic bands of ^{51}Cr-labeled oligomers. To avoid this, the Th^{4+} cation was replaced with Ce^{4+} by using a saturated solution of $(NH_4)_4Ce(SO_4)_4$. Once the trimer band was eluted with La^{3+}, a concentration gradient of Ce^{4+} was used to remove the tetramer and other oligomers from the column. These are labeled in Figure 1B. Interestingly, this gave quantitative recovery of all of the Cr(III) from the column, as shown in Table 1. The total volume required for complete elution was large compared to the separation using Th^{4+} (Figure 1A), and complete resolution of many of the bands was not achieved. The separation was improved (Figure 1C) by increasing the volumes used for all the eluants, and a distinct tetramer fraction was isolated. This fraction was followed by bands containing species that are likely larger than tetrameric. However, these species could not be identified, because the electronic spectra, stoichiometry, and structures of such species are not defined.

Capillary Electrophoresis. In CE, separation is based on net charge density rather than simply cationic charge so that the order of elution for a given set of species such as shown in Chart 1 is not always easily predictable a priori. To uniquely identify the species represented by the various chromatographic bands in an electropherogram, a process of elimination using the Cr(III) oligomer fractions we isolated by ion exchange was employed. Even with efforts to remove or reduce the concentrations of eluant cations used in the ion exchange, these species are still present, each of which is also separated into distinct bands in CE. The order of elution of the simple cations has been reported by others,[22] and similar results are shown in Figure 2A. This solution also contained monomeric Cr(III), as indicated. Figure 2B is an electropherogram of the dimer fraction obtained by ion exchange. Note that two Cr(III) species are present. To uniquely identify each, a spike of monomeric Cr(III) was added to the dimer solution, and the resulting electropherogram is also shown. The peak area that increased was identified as the monomer, and by default, the other peak was identified as the dimer. Relative retention times for the trimer and tetramer were obtained by injecting isolated fractions of each species separately, as shown in Figure 2C,D. Absolute retention times for each species in the different electropherograms cannot be directly compared because

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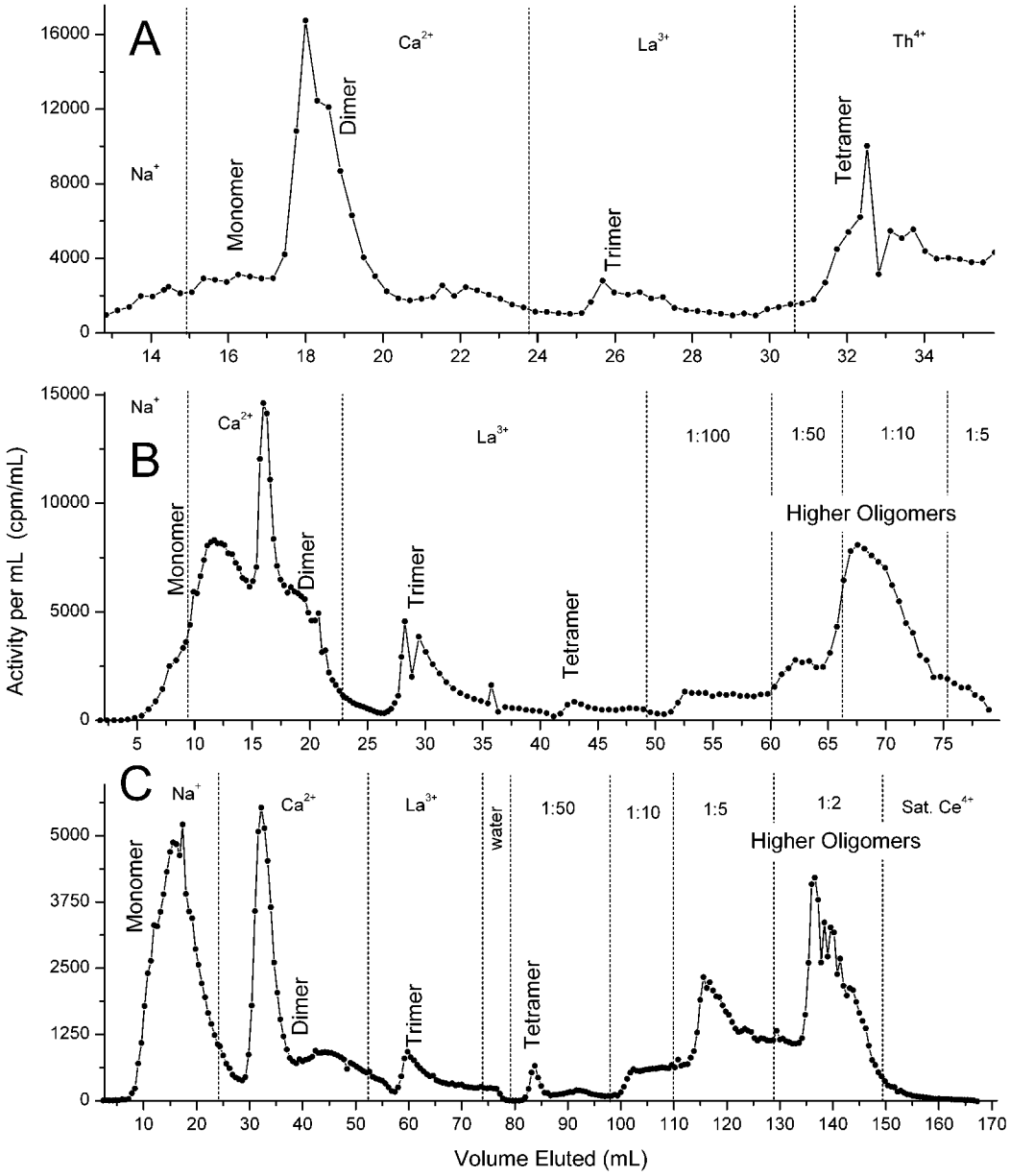

Figure 1. Chromatograms for the elution of ^{51}Cr-labeled Cr(III) oligomers by ion exchange. Note the differing scales on the *x* axes. Sephadex SP C-25 served as the cation exchanger; columns were 1-cm i.d. with 8 cm of resin. Total Cr(III) concentration was 1×10^{-2} M. (A) Elution was with perchlorate-based solutions of Na^+, Ca^{2+}, La^{3+}, and Th^{4+}, as shown. More than 44% of the Cr(III) remained on the column after completion of the separation. Quantification of this "irreversibly sorbed" Cr(III) is described in Table 1. (B) Elution with perchlorate-based solutions of Na^+, Ca^{2+}, La^{3+}, followed by various dilutions of saturated $(NH_4)_4Ce(SO_4)_4$, as indicated. Dilutions of the Ce^{4+} solutions were made with 18 MΩ H_2O. Upon completion of the separation, ~2% of the Cr(III) remained on the column (Table 1). Two 5.0-mL washes with perchloric acid removed most of the sorbed Cr(III), as shown in Table 1. (C) A second chromatogram obtained by elution with perchlorate based solutions of Na^+, Ca^{2+}, La^{3+}, followed by various dilutions of saturated $(NH_4)_4Ce(SO_4)_4$, as indicated. Larger volumes of eluants were used to improve resolution between the bands of Cr(III) oligomers, compared to Part B. This approach left very little Cr(III) on the column (see Table 1).

of differing operating conditions, but the retention times relative to the EOF are comparable, and the elution order for the species is always the same.

A typical electropherogram for the Cr(III) species present in a hydrolyzed solution that had not been separated by ion exchange is shown in Figure 3. Because no prior separation was attempted with this solution, no other metal cations are present except for K^+ from the base. The smaller Cr(III) oligomers that we could uniquely identify are marked; in addition, other Cr(III) species of higher net charge density are present but cannot be uniquely identified. These species are clearly important and ideally should be considered in Cr(III) speciation and solubility work. Unfortunately, lack of stoichiometric and structural information on these species limits their consideration. CE can also be used to check the purity of the Cr(III) oligomers isolated by ion exchange. Although spectrophotometric and radiometric data indicates that complete peak resolution was achieved by our ion-exchange methods (e.g., Figure 1C), electropherograms of each fraction collected as soon as possible (~24 h) after separation indicates that other Cr(III) species are present. For example, an electropherogram of the dimer solution after ion-exchange separation showed that monomer was present (e.g., Figure 2B). Similarly,

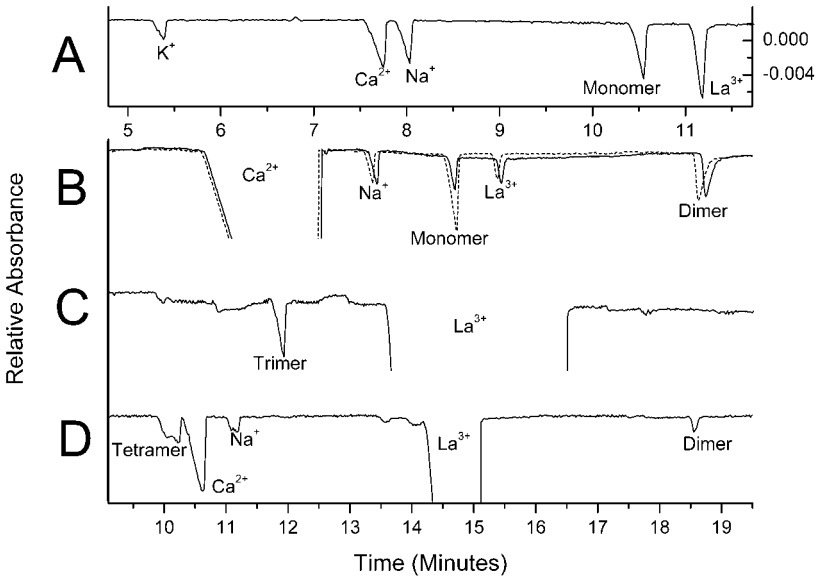

Figure 2. Electropherograms obtained by completing capillary electrophoresis using indirect detection on the Cr(III) oligomer fractions separated by ion exchange. The buffer used was α-hydroxyisobutyric acid/dimethylbenzylamine (HIBA/DBA), as described in the Experimental Section. (A) CE analysis of a solution containing the simple cations used as eluants and the Cr(III) monomer. The concentration of ions present is ~1 × 10^{-4} M. Voltage was 30 000 V. (B) Electropherogram for the dimer fraction. The solid line is for the dimer fraction obtained by ion exchange. The dashed line is the same solution to which a spike of monomeric Cr(III) was added. Voltage was 20 000 V. (C) CE analysis of the trimer fraction using 20 000 V. (D) CE separation of the tetrameric Cr(III) fraction at 20 000 V.

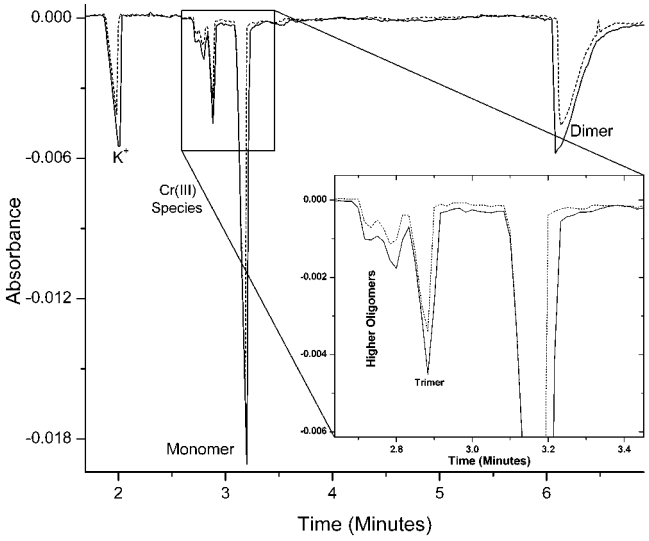

Figure 3. Electropherogram for a hydrolyzed solution of Cr(III). This solution had not been subjected to ion exchange. The bands for the Cr(III) species are labeled. $[Cr(III)]_{total}$ = 5.0 × 10^{-3} M, HIBA/DBA buffer, total column length was 22 cm, voltage was 30,000 V.

Table 2. Estimated Volumes and Charge Densities for the Various Cr(III) Species Shown in Chart 1[a]

	linear species		nonlinear species	
	V (Å^3)	charge density	V (Å^3)	charge density
monomer	284.5	0.0105		
dimer	469.9	0.00851		
trimer	655.3	0.00763	559.5	0.00894
tetramer	840.8	0.00714	649.1	0.00924

[a] Details on the calculations used to obtain the volume estimates are provided in Supporting Information. The charge densities were obtained by dividing the charge of the species by its volume.

the tetrameric fraction always indicated traces of dimer. The smaller Cr(III) species must be the result of dissociation of the larger oligomers with time, consistent with LaChatlier's principle of equilibrium.

For those Cr(III) species that can be uniquely identify in the electrophrograms, some structural information can be inferred. For example, using formulas 1 and 2, we can calculate the size of the Cr(III) monomer from its retention time in CE and estimate its hydrated radius to be ~4.69 Å. This is somewhat large compared to 4.08 Å, estimated by extended X-ray absorption fine structure (EXAFS) analysis of the Cr(III) monomer in aqueous solution.[24] The EXAFS results include a second shell of water molecules, suggesting an extended hydrated radius, but no interactions beyond the second shell of water molecules could be observed. Note that tertiary hydration shells are believed to exist for the trivalent lanthanide cations in aqueous solutions (e.g., see ref 25) which would increase the apparent size of the Cr(III) monomer; however, the possibility of interaction between the Cr(III) monomer and the HIBA electrolyte, as reported for other metal cations, cannot be excluded.[21] In addition, although others have not been able to exclude the possibility of linear configurations for the larger Cr(III) oligomers (especially for the trimer[7]), the order of elution we observe in CE suggests that these species do not exist to any significant extent. As shown in Table 2, the EXAFS information is used to estimate volumes and charge densities for the various species shown in Chart 1. (For details on these calculations, see Supporting Information.) For the linear species, the estimated cationic charge densities decrease with increasing oligomerization, which suggests that the elution order should be monomer, followed by dimer, followed by trimer, followed by tetramer. However, the tetramer and trimer elute

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before the monomer and dimer. This demonstrates that the trimeric and tetrameric species have larger net charge densities than the monomeric and dimeric species; such charge densities are not likely for the linear configurations. Therefore, the trimer and tetramer likely exist as the nonlinear conformers shown in Chart 1C and E, respectively. This is consistent with the work of Stunzi et al.,[9] who used pK_a measurements for the μ-hydroxo protons to indirectly support the predominance of the nonlinear Cr(III) trimer.

CONCLUSIONS

We have shown that specific oligomers of Cr(III) can be isolated in concentrated fractions by ion exchange using solutions of simple cations of increasing positive charge as eluants. With sufficient diligence, separation of hydrolyzed species of other charged metal cations such as Fe(III), Al(III), and Pu(IV) should also be possible, and ion exchange provides sufficient volumes and concentrations for other subsequent analyses, such as spectroscopic characterization. CE using indirect detection affords simpler, microscale separations, but unique characterization of the isolated species requires additional work. Whereas we used a process of elimination based on our ion-exchange work, other approaches could include coupling CE with mass spectrometry, as demonstrated by others.[26−29] The chromatographic separations we described herein are necessary for improving modeling calculations in solution speciation and solubility studies for the higher metal valent cations and for studying the rates and mechanisms by which metal cations hydrolyze, nucleate, and eventually precipitate. In addition, our results demonstrate that the reverse processes (e.g., dissociation of larger Cr(III) oligomers into smaller Cr(III) species) are also possible and provide pathways for ingrowth of smaller oligomers. Study of these processes has been inhibited by lack of analytical tools, such as effective methods to separate and identify the important species that are present. Although our chromatographic separations reported herein will aid in these studies, additional spectroscopic work is necessary to determine the structures of the higher Cr(III) oligomers.

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SUPPORTING INFORMATION AVAILABLE

Details on the calculations used to obtain volume estimates and charge densities for the various species shown in Chart 1. This material is available free of charge via the Internet at http://pubs.acs.org.

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